

Project Title: Microbiological Impact of Agricultural and CAFO Activities on Surface Water Quality

Principal Investigators: Y. Olivas

Project Period: 04/01/2002 - 09/30/2005

Long-Term Goal/APM: Long Term Goal 3 - Provide the tools to restore and protect aquatic systems and to forecast the ecological, economic, and human health outcomes of alternative solutions/APM 144 (2004) - Develop a strategy to evaluate BMP performance via molecular based methods in watersheds impaired by fecal contamination (*contributes to this APM*)

Abstract: Previous evidence of microbial contamination of natural waters by agriculture and Concentrated Animal Feeding Operations (CAFOs) has created the necessity to further evaluate the situation. Pathogenic bacteria and pathogenic viruses are often present in the intestinal microflora of humans, as well as animals associated with CAFOs. This creates the potential for pathogen transfer into nearby watersheds, which in turn poses a public health threat. The objective of this research is to provide a microbiological evaluation of the Turkey Creek Watershed located in northwest central Oklahoma. This is a priority watershed listed under Section 303D of the Clean Water Act, and is currently the focus of a USGS study on sources of impairment. Nitrogen-isotope ratios of nitrate and detection of organic compounds typical of human wastewater previously indicated that animal and/or human wastes were the major sources of nitrate in Turkey Creek. Therefore, it is important to identify the species source of fecal contamination as well as specific pathogens that may be present in the watershed. This research project consists of two phases. The first phase is dedicated to bacterial source tracking using the bacterium *E. coli* as the indicator organism. The methodology is based on antibiotic resistance analysis in combination with a statistical discriminant analysis. The second phase will involve identification of specific viral and bacterial pathogens in the water. The target microorganisms of this project are the human enteroviruses (poliovirus, coxsackievirus, echovirus) and the bacteria *Yersinia enterocolitica*, and *Campylobacter jejuni*. The methodology will be based on hollow-fiber ultra-filtration and real-time PCR technology. The application of the results will aid in the development of TMDLs and risk management strategies for optimizing land use practices in the animal industry.

Status: The first phase of this project is almost complete. The field work and analytical recovery of the bacterium *E. coli* have been done, and an analysis of seasonal and spatial fecal coliform trends along the stream has been completed. A total of 3600 source-isolates and 3000 water-isolates have been recovered and stored for antibiotic resistance analysis, which is in progress and is planned to be completed by July 2004. Preparative work on the second phase will begin Jan 2004 to develop pathogen recovery and PCR procedures, and recovery and identification work will commence Aug 2004.

Products: (1) Paper on sources of coliforms in a TMDL-listed watershed (*planned*), (2) paper on recovery and identification of pathogens in a TMDL-listed watershed (*planned*).